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Sacubitril/valsartan: Beyond natriuretic peptides

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Abstract

Natriuretic peptides, especially B-type natriuretic peptide (BNP), have primarily been regarded as biomarkers in heart failure (HF). However, they are also possible therapeutic agents due to potentially beneficial physiological effects. The angiotensin receptor-neprilysin inhibitor (ARNI), sacubitril/valsartan, simultaneously augments the natriuretic peptide system (NPS) by inhibiting the enzyme neprilysin (NEP) and inhibits the renin-angiotensin-aldosterone system (RAAS) by blocking the angiotensin II receptor. It has been shown to improve mortality and hospitalisation outcomes in patients with HF due to left ventricular systolic dysfunction. The key advantage of sacubitril/valsartan has been perceived to be its ability to augment BNP, while its other effects have largely

been overlooked. This article highlights the important effects of sacubitril/valsartan, beyond just the augmentation of BNP.

First we discuss how NPS physiology differs between healthy individuals and those with HF by looking at mechanisms like the overwhelming effects of RAAS on the NPS, natriuretic peptide receptor desensitisation and absolute natriuretic deficiency. Secondly, this review explores other hormones that are augmented by sacubitril/valsartan such as, bradykinin, substance-P and adrenomedullin that may contribute to the efficacy of sacubitril/valsartan in HF. We also discuss concerns that sacubitril/valsartan may interfere with amyloid β homeostasis with potential implications on Alzheimer's disease and macular degeneration. Finally, we explore the concept of 'auto-inhibition' which is a recently described observation that humans have innate NEP inhibitory capability when natriuretic peptide levels rise above a threshold. There is speculation that auto-inhibition may provide a surge of natriuretic and other vasoactive peptides to rapidly reverse decompensation. We contend that by pre-emptively inhibiting NEP, sacubitril/valsartan is inducing this surge earlier during decompensation, resulting in the better outcomes observed.

Introduction

Sacubitril/valsartan is the first in a new class of drug: the angiotensin receptor - neprilysin inhibitors (ARNI). Its mechanisms of action have not been well defined. Sacubitril/valsartan causes simultaneous augmentation of the natriuretic peptide system (NPS) and inhibition of the renin-angiotensin-aldosterone system (RAAS).[1] RAAS inhibition by valsartan has been extensively tested in patients with heart failure (HF).[2] Therefore, in this article we shall focus on the actions of sacubitril, which are less well understood.

The Prospective Comparison of ARNI [Angiotensin Receptor–Neprilysin Inhibitor] with ACEI [Angiotensin-Converting– Enzyme Inhibitor] to Determine Impact on Global Mortality and Morbidity in Heart Failure Trial (PARADIGM-

HF) trial,[3] which was stopped early due to overwhelming benefit, reported an impressive 20% relative risk reduction (4.7% absolute risk reduction) in the primary outcome of HF hospitalisation or cardiovascular (CV) death with sacubitril/valsartan when compared to enalapril. The beneficial effect of sacubitril/valsartan has been perceived to be due to its effect on the NPS, while its other effects have largely been overlooked. This has led some to think that increasing the already elevated natriuretic peptide hormones in heart failure even further will be of little benefit. Indeed, trials testing other methods of modulating the NPS in acute HF, such as by supplementing exogenous natriuretic peptides, direct NEP inhibition or dual inhibition of NEP and ACE, have been disappointing. (Table 1)

The purpose of this review is to understand the effects of sacubitril/valsartan on the NPS and other vasoactive systems, and to explore alternate mechanisms that account for not only the beneficial effects, but also possible off-target effects seen with sacubitril/valsartan.

The Natriuretic Peptide System

There are a number of natriuretic peptides that play an important role in CV homeostasis, namely atrial natriuretic peptide (ANP), B-type natriuretic peptide (BNP), C-type natriuretic peptide (CNP), dendroaspis-type natriuretic peptide (DNP), and urodilatin.[4] ANP and BNP are predominantly produced by the atria

and ventricles respectively in response to volume/pressure overload.[4] (Figure 1) They both increase glomerular filtration rate, and enhance sodium and water excretion in the kidneys.[5] They also promote vasodilatation, antagonise vasoconstriction and increase capillary permeability, causing fluid loss into the extravascular compartment.[6] Additionally, natriuretic peptides inhibit the secretion of renin and aldosterone, and antagonise the sympathetic system.[6]

All natriuretic peptides act on two guanylate cyclase-linked transmembrane receptors, natriuretic peptide receptor (NPR) A and B. NPR-A primarily binds to ANP and BNP, whereas NPR-B has a higher affinity for CNP.[7] When activated the guanylate cyclase moiety within the receptor catalyses the production of second messenger cyclic guanosine monophosphate (cGMP) from guanosine triphosphate (GTP). This cGMP pool then mediates a variety of downstream signalling cascades in the target organs (vascular endothelium, cardiac myocytes and fibroblasts, zona glomerulosa of the adrenal cortex and renal epithelial cells).[7] (Figure 2)

Unlike most other receptor classes, NPR-A and NPR-B are single-molecule trans-membrane receptors that do not require internalisation when a ligand binds.[6] Consequently, they cannot be down-regulated.[8] Nevertheless, long term exposure to high concentrations of natriuretic peptides results in receptor *desensitisation*

secondary to dephosphorylation,[8] meaning the receptor concentration and binding capacity remain unchanged but the receptors are unable to generate the same second messenger response as before.

NPR-C, functions as a clearance receptor. It binds to all natriuretic peptides and internalises them for lysosomal hydrolysis.[9] Unlike NPR-A and NPR-B, it has three distinct domains (extracellular, transmembrane and intracytoplasmic) and can therefore undergo down-regulation. NPR-C is the most abundant class of NPR, comprising more than 95% of the total NPR population, and is thus the dominant mode of natriuretic peptide clearance.[6]

An alternative metabolic pathway for natriuretic peptides is hydrolysis by a membrane bound metalloproteinase called neprilysin or neutral endopeptidase 24.11 (NEP). It is distributed widely (brain, eyes, lungs, intestines, fibroblasts) but is predominantly found in the brush border of proximal tubular cells in the kidneys.[10] NEP metabolism is a minor contributor to natriuretic peptide clearance under normal conditions but becomes the dominant clearance pathway in disease states such as HF, when clearance via the NPR-C pathway becomes saturated (or down-regulated following chronic exposure).[11]

The Natriuretic Peptide Paradox of Heart Failure

The mechanisms discussed above describe the function and action of the NPS in isolation. In a healthy individual, these mechanisms hold true, but in HF a very different picture emerges. In chronic HF, patients develop a state of resistance to natriuretic peptides,[12] such that despite persistently elevated natriuretic peptide levels, patients remain congested. This then raises the question: why would increasing natriuretic peptides further by using a drug like sacubitril/valsartan be beneficial? To answer this fundamental question, we will need to first understand the pathophysiology behind this 'natriuretic peptide paradox'.[13] A number theories have been proposed, and the true mechanism behind this phenomenon may well be a combination of the following theories:

a. Overwhelming effects of the RAAS on the actions of natriuretic peptides

In healthy individuals, there is an inverse relationship between ANP and renin suggesting the NPS and RAAS are mutually antagonistic. However, this relationship is lost in patients with HF, who exhibit a positive correlation instead.[14] This reflects the simultaneous activation of both the NPS (as a result of atrial distension and increased ventricular end diastolic pressure) and RAAS (due to reduced blood pressure and renal perfusion pressure).[15] Even though most of the effects of the NPS are opposed to the RAAS, the RAAS is capable of overwhelming the NPS; angiotensin II (Ang II) attenuates the natriuretic effect

of ANP in the kidneys of healthy individuals.[16] Additionally, angiotensin and vasopressin desensitise vascular ANP receptors in the rat, perhaps via protein kinase C activation, thereby suppressing NPR-A second messenger cGMP production.[17]

b. Receptor desensitisation:

Tsutamoto et al. demonstrated that patients with chronic HF had higher ANP extraction (i.e. binding of ANP to its receptor) but lower cGMP (second messenger) production than those with acute HF. This suggests that although patients with chronic HF have more receptors, they were unable to produce the same amount of cGMP. More importantly, they also demonstrated that it was the *duration* of HF and not the severity of HF that determined receptor desensitisation.[12] The mechanism behind the desensitisation of chronically ligand-bound natriuretic peptide receptors has been attributed to the dephosphorylation of their kinase domains. This prevents the receptors from undergoing the conformational change required to activate the guanylate-cyclase moiety responsible for the conversion of GTP to cGMP.[18] (Figure 2)

c. Natriuretic peptide deficiency:

Some studies have suggested that the predominant portion of circulating BNP in patients with advanced HF is unprocessed proBNP (BNP_{1-108}), and not the biologically active BNP (BNP_{32}), suggesting that patients actually have a state of natriuretic peptide deficiency;[19] in other words, the myocardium continues to produce the natriuretic peptide prohormones in response to the volume and pressure stressors of HF, but these prohormones are not being processed into their biologically active forms.

Corin is the cardiac transmembrane serine protease which is the putative major convertase enzyme for both proANP and proBNP.[20] Some studies have suggested that patients with advanced heart failure are unable to mount a natriuretic response because of corin dysregulation, resulting in unprocessed proBNP (BNP_{1-108}) being the predominant portion of circulating BNP, instead of the biologically active BNP (BNP_{32}). (Figure 1) In a small study using novel immunoassays, patients presenting with acute HF had plasma corin levels less than one eighth of those in healthy individuals.[21] Plasma corin levels were also lower among patients with chronic HF compared to healthy controls, and were inversely correlated with the severity of heart failure.

Additionally, commercial antibody-based enzyme-linked immunosorbent assay (ELISA) detection methods are unable to distinguish between unprocessed BNP_{1-}

$_{108}$, active BNP_{32} or fragments of the two (e.g. BNP_{1-30}, BNP_{3-32}, BNP_{4-32} and BNP_{5-32}). Among HF patients, mass spectrometry analysis suggests that the true levels of BNP_{32} (active BNP) were significantly lower than those measured by ELISA.[20] The ELISA-based BNP assays cross-react with BNP_{1-108} and its breakdown fragments leading to spuriously high measured BNP levels in patients who are actually (BNP_{32}) deficient.[22]

Pharmacology of sacubitril/valsartan

Sacubitril/valsartan is a novel compound that combines sacubitril and valsartan in a salt delivering a 1:1 molar ratio of its constituents after oral administration.[23] Valsartan is an angiotensin receptor blocker with proven efficacy in cardiovascular disease.[24] Sacubitril is a prodrug and is metabolised to sacubitrilat (LBQ 657), which then inhibits NEP.[23] With the mean elimination half-lives of sacubitril, sacubitrilat and valsartan being 1.4 hours, 11.5 hours and 9.9 hours respectively,[23] sacubitril/valsartan is suitable for once daily administration, although it is used as a twice daily preparation in heart failure to ensure a sustained and uninterrupted effect on both NPS and RAAS.[25]

The direct consequence of NEP inhibition is an increase in circulating natriuretic and other vasoactive peptides. NEP does not break down natriuretic precursor molecules such as proBNP or its N-terminal (NT) fragment, and their plasma levels are therefore not directly affected by NEP inhibition.[26] ProBNP and NT-

proBNP thus remain useful biomarkers in patients treated with sacubitril/valsartan. (Figure 3) In fact, NT-pro-BNP is indirectly reduced by sacubitril/valsartan in HF as a consequence of reduced ventricular wall stress.[27]

Sacubitril/valsartan and its effects on other peptide hormones

It is unlikely that the benefits reported in the PARADIGM-HF trial are attributable solely to inhibition of RAAS and potentiation of NPS by sacubitril/valsartan. NEP is a 'promiscuous enzyme' with a long list of potential substrates including enkephalins, oxytocin, gastrin, Ang I and II, endothelin-1, adrenomedullin, substance P, and bradykinin[25] (each with its own kinetics). In vitro studies show that NEP has a far greater affinity for some of these peptides than for BNP (Table 2), suggesting that other peptides may be involved (at least in part) in producing the beneficial effects reported in PARADIGM-HF. (Figure 4) The effects of sacubitril on other peptides have not been fully explored in humans, however there is animal data that signal potential benefits. .

Substance P and bradykinin have frequently been blamed for such adverse effects of ACE inhibitors as dry cough and angioedema. In clinical trials testing omapatrilat (a drug that simultaneously inhibits ACE and NEP), there was marked increase in angio-oedema.[1] This was attributed to excessive potentiation

of bradykinin and substance P levels due to the inhibition of ACE, NEP and aminopeptidase P.[1] Nevertheless, substance P and bradykinin have also shown potential CV benefit. [28 29]

a. Substance P

Substance P is a vasoactive neuropeptide that is also found in the human heart including the adventitia of the coronary vessels[30] and between the cardiomyocytes.[31] It is secreted by endothelial cells in response to sheer stress to produce vasodilatation in the coronary vasculature by binding to neurokinin (NK)-1 receptors, which releases nitric oxide.[32]

Animal ischaemia reperfusion studies show substance P having a protective effect by increasing coronary perfusion and attenuating hypoxic cellular damage.[33] However chronic exposure to substance P induces inflammation, apoptosis and matrix metalloproteinase activation which result in adverse remodelling.[33]

b. Bradykinin:

Bradykinin is another potent vasodilatory peptide that acts via B2 kinin receptors in the vascular endothelium to stimulate the synthesis of nitric oxide, prostacyclin and endothelium-derived hyperpolarising factor resulting in vasodilatation.[34] It

preferentially increases blood flow to the subendocardium,[34] thus improving transmural myocardial perfusion. Increased bradykinin levels due to ACE-inhibition result in reduced renal vascular resistance due to selective efferent arteriolar dilatation.[35] The salutary effects of bradykinin potentiation as a result of ACE inhibition is seen in in patients with[36] and without HF.[37]

c. Adrenomedullin

Adrenomedullin is synthesized in a variety of tissues including the adrenal glands, endothelium, vascular smooth muscles, renal parenchyma and cardiac myocytes. It has multiple potentially beneficial effects such as vasodilatation, anti-proliferation, increased renal blood flow, natriuresis and diuresis.[38] Clinical studies show that adenomedullin reduces pulmonary capillary wedge pressure, increases cardiac index and increases urinary volume and sodium excretion in patients with HF.[39]

Effects of sacubitril/valsartan on amyloid β

The pathophysiology of Alzheimer's disease (AD) is complex. The leading hypothesis behind AD suggests late onset AD (the commonest type of AD) to be the sequelae of reduced amyloid β clearance.[40] There are two mechanisms for amyloid β clearance, namely enzymatic or non-enzymatic (transport proteins)

clearance. NEP is involved in the enzymatic clearance of amyloid β. Clinical studies show declining expression of NEP in the hippocampus and midtemporal gyrus of patients with AD in parallel with increasing deposition of amyloid plaques,[41] while areas that are resistant to amyloid plaque deposition, such as the caudate nucleus, show increased NEP expression.[42]

There was no increase in the risk of dementia or cognitive decline over the median follow-up time of 27 months in the patients treated with sacubitril/valsartan in PARADIGM-HF.[43] In young cynomolgus monkeys, sacubitril/valsartan resulted in increased amyloid β in the cerebrospinal fluid (CSF) but there were no amyloid plaques in the brain parenchyma.[23] A study on healthy volunteers over 2 weeks showed that sufficient concentration of sacubitrilat was achieved in the CSF to inhibit NEP in the brain. There was an increase in the amyloid β1-38 isoform which is hydrophilic and does not aggregate to form amyloid plaques. Levels of amyloid β1-40 and β1-42 concentrations in the CSF (which *do* form amyloid plaques) were unchanged from baseline.[44] These and other pre-clinical studies[45] seem to suggest that although NEP inhibition in the brain results in a net increase in total amyloid concentration, the increase is primarily driven by the soluble, non-plaque forming, amyloid β1-38 isoform. Further evaluation is required as these observations were made over the short term in young and healthy subjects.

As a result of the possible risk of AD, there is now a trial investigating the effects of sacubitril/valsartan compared to valsartan on cognitive function in patients with HF. As part of the trial, the US Food and Drug Administration (FDA) has suggested a comprehensive battery of neurocognitive tests as well as positron emission tomography studies. (Clinical trials identifier: NCT02884206) Results are expected in 2022.[46]

In the eye, amyloid β deposits have been linked to age-related macular degeneration (AMD). NEP deficient mice develop retinal pigment epithelial cell degeneration and sub-retinal deposits that result in AMD.[47] This is reversed when the catalytic domain of NEP is administered into the vitreous humour of a mouse model of retinal degeneration.[48] There are no published data on the effects of sacubitril/valsartan on the eye in humans.

Auto-inhibition of neprilysin

Vodovar and colleagues studied 684 patients, 468 of whom had decompensated HF.[49] They found that raised BNP (above 916 pg/ml) was associated with reduced circulating NEP activity. They then incubated the plasma of age-matched healthy controls with either high or low concentrations of recombinant BNP. NEP *activity*

was much lower in the plasma incubated with high levels of BNP, even though the NEP *concentration* was the same in both groups.

These data suggest that BNP acts as a 'molecular switch', high concentrations of which can inhibit the activity of NEP thereby inducing a further accumulation of natriuretic peptides and other vasoactive peptides that are substrates of NEP (including substance P, bradykinin and adrenomedullin).[49]

This phenomenon of NEP 'auto-inhibition' by BNP raises a theoretical possibility that sacubitril/valsartan will have little benefit beyond that of angiotensin receptor blockade (conferred by the valsartan component) in patients with advanced HF and very elevated BNP (who consequently will have NEP auto-inhibition), rendering the sacubitril component redundant. Although recent post-hoc analysis of the PARADIGM-HF dataset showed the benefit of sacubitril/valsartan was seen regardless of baseline BNP levels,[50] the only way this question can reliably be answered is by a clinical trial. Indeed, the LIFE-HF trial (NCT02816736) will be attempting to answer this question.

Pre-empting HF decompensation

In a patient with compensated HF, NPR-C is the dominant clearance pathway for the NPS. However, when the patient decompensates and starts to produce large

quantities of natriuretic peptides, the NPR-C receptors become saturated and the NEP pathway becomes the primary pathway for clearance.[6] In a rat model, inhibition of NEP alone does not affect the plasma half-life of ANP. However, blocking NPR-C (thereby simulating the NPR-C saturation seen in HF) doubles the half-life of BNP, and blockade of both NEP *and* NPR-C prolongs it further.[11] Sacubitril thus probably has little effect in patients with compensated HF because in that state NEP is only a minor metabolic pathway.

A New Paradigm in Heart Failure

It is important to remember that compensated HF and (acute) decompensated HF are two ends of the same disease spectrum. The discussion above regarding NEP auto-inhibition and NPR-C clearance seem to suggest that sacubitril/valsartan would probably be most effective in the area between the two ends of the HF spectrum.

When a stable patient already on sacubitril/valsartan starts to decompensate (thereby saturating their NPR-C receptors), the pre-emptively inhibited NEP starts to have a multiplicative effect on the rising levels of its substrates, as illustrated in figure 5. This results in a surge in the activity of not only the NPS but also other vasoactive hormones such as substance P, bradykinin and adrenomedullin. This early and pronounced burst of activity has

haemodynamically beneficial effects (such as coronary and systemic vasodilatation, diuresis, reduced sympathetic activity), thereby arresting the decompensation. As a result of fewer episodes of decompensation, we can expect to delay the progression of heart failure. (Figure 5)

It may well be that this ability to dynamically modulate the natriuretic and vasoactive peptide systems is what sets sacubitril/valsartan apart from the other HF therapies that have more static pharmacodynamic profiles which are unable to respond as the patient moves along the HF spectrum.

Conclusions

With recent approvals from the FDA, the European Medicines Agency and the National Institute for Health and Care Excellence, sacubitril/valsartan is poised to change the way we treat patients with HF. It is unlikely that all the benefits of sacubitril/valsartan can be explained simply by its effects on the NPS or BNP alone. The complete mechanism of benefit of sacubitril/valsartan is not yet fully elucidated and only with more rigorous research will it become clear. It is imperative that we understand how this drug affects the various hormonal pathways, not only to ensure its appropriate use and to recognise its limitations, but also to help guide the development of newer therapies with other targets.

Table 1. Comparison table of trials testing treatment that modulate the natriuretic peptide system.

Footnote: VMAC= Young et al. Vasodilation in the Management of Acute Congestive Heart Failure. *Circulation* 2000;102:2794. **ASCEND-HF**= O'Connor et al. Effect of nesiritide in patients with acute decompensated heart failure. *The New England journal of medicine* 2011;365(1):32-43. **META-ANALYSIS=** Sackner-Bernstein et al. Risk of worsening renal function with nesiritide in patients with acutely decompensated heart failure. *Circulation.* 2005;111(12):1487-1491. **U.S. Ecadotril Pilot Safety Study**= O'Connor et al. A randomized trial of ecadotril versus placebo in patients with mild to moderate heart failure: the US ecadotril pilot safety study. *Am. Heart J*. 138,1140–1148. **International, Multicentre Ecadotril Dose-ranging Study**=Cleland et al. Lack of efficacy of neutral endopeptidase inhibitor ecadotril in heart failure. The International Ecadotril Multicentre Dose-Ranging Study Investigators. *Lancet* 351, 1657–1658. **Northridge et al.=** Northridge et al. Placebo-controlled comparison of candoxatril, an orally active neutral endopeptidase inhibitor, and captopril in patients with chronic heart failure. *European Journal of Heart Failure* 1999;1(1):67-72. **Westheim et al.**= Westheim et al. Hemodynamic and neuroendocrine effects for candoxatril and frusemide in mild stable chronic heart failure. *Journal of the American College of Cardiology.* 15 Nov 1999;34(6):1794-1801. **Kentsch et al.=** Kentsch et al. Neutral endopeptidase 24.11 inhibition may not exhibit beneficial haemodynamic effects in patients with congestive heart failure. *European journal of clinical pharmacology* 1996;51(3-4):269-272. **IMPRESS**= Rouleau et al. Comparison of vasopeptidase inhibitor, omapatrilat, and lisinopril on exercise tolerance and morbidity in patients with heart failure: IMPRESS randomised trial. *Lancet.* 2000;356(9230):615-620. **OVERTURE**= Packer et al. Comparison of omapatrilat and enalapril in patients with chronic heart failure: the Omapatrilat Versus Enalapril Randomized Trial of Utility in Reducing Events (OVERTURE). *Circulation.* 2002;106(8):920-926. **OCTAVE**= Kostis et al. Omapatrilat and enalapril in patients with hypertension: the Omapatrilat Cardiovascular Treatment vs. Enalapril (OCTAVE) trial. *American journal of hypertension.* 2004;17(2):103-111. **PARADIGM-HF**= McMurray et al. Angiotensin-neprilysin inhibition versus enalapril in heart failure. *The New England journal of medicine.* 2014;371(11):993-1004. **PARAMOUNT**= Solomon SD, Zile M, Pieske B, et al. The angiotensin receptor neprilysin inhibitor LCZ696 in heart failure with preserved ejection fraction: a phase 2 double-blind randomised controlled trial. *Lancet.* 2012;380(9851):1387-1395. **PARAGON=** Clinical trials identifier: NCT01920711.

Table 2. Peptide substrates of neprilysin.[51]

Footnote: Km=Michaelis constant - the concentration of a substrate which allows the enzyme to achieve half of maximum reaction velocity, it is the inverse measure of affinity of an enzyme to its substrate (ie. lower the Km, the higher the affinity of an enzyme to the substrate); Kcat=catalytic production rate under optimum conditions; Kcat/Km= comparison ratio that allows for evaluation of the efficiency of an enzyme on different substrates

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JSSS, LMB, MC, IBS, ALC & CCL co-developed the hypothesis and drafted the manuscript.

Disclosures

JSSS reports no conflicts of interest.

LMB has received consulting fees from Novartis and lecture fees from AstraZeneca.

MC is an employee of Novartis Pharmaceuticals UK Limited.

IBS has received fees from Novartis for participation in advisory boards and educational events.

ALC Andrew Clark has received financial support for travel to meetings from Servier, and has accepted fees for service on behalf of his department from Novartis.

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FIGURE LEGENDS

Figure 1. The stimulus for and sites of synthesis of ANP, BNP and CNP.

ANP=Atrial natriuretic peptide; BNP=B-type peptide; CNP=C-type peptide; NT=N-terminal; LA=left atrium; RA=right atrium; LV=left ventricle; RV=right ventricle; EDP=end diastolic pressure;

Figure 2. The mechanism of action and the pathways of metabolism of ANP, BNP and CNP.

ANP=Atrial natriuretic peptide; BNP=B-type peptide; CNP=C-type peptide; NPR=natriuretic peptide receptor; NEP=neprilysin, GTP=guanosine triphosphate; cGMP=cyclic guanosine monophosphate, CV=cardiovascular

Figure 3. Schematic representation of mechanism of sacubitril/valsartan on the natriuretic peptide and renin-angiotensin-aldosterone systems.

BNP= B-type natriuretic peptide; Na= Sodium; ACE=angiotensin converting enzyme

Figure 4. Overview of pathophysiological effects of HF and the multi-modal mechanisms of sacubitril/valsartan in reversing those effects. Detrimental effects coloured grey, beneficial effects of sacubitril/valsartan coloured blue (NPS) and red (RAAS).

NPS=natriuretic peptide system; NP= natriuretic peptide; ANP=atrial natriuretic peptide; BNP=B-type natriuretic peptide; CV=cardiovascular; BP= blood pressure; RAAS=renin-angiotensin-aldosterone system; AT=angiotensin; ?=unknown or theoretical effect

Figure 5. Hypothetical excursions of natriuretic and other vasoactive peptides (eg bradykinin, substance P and adrenomedullin) during episodes of acute

decompensated heart failure (blue line) and baseline (compensated) BNP levels (red line).

Figure 5a. Patients not on sacubitril/valsartan gradually increase natriuretic and vasoactive peptide levels in response to decompensation. This delays the onset of the beneficial effects of these peptides that can only manifest after overcoming natriuretic peptide receptor desensitisation and the counter-regulatory effects of RAAS.

Figure 5b. Patients on sacubitril/valsartan will already have neprilysin inhibition prior to decompensation. When an episode of decompensation occurs, natriuretic and vasoactive peptide levels rise rapidly and begin to manifest their effects earlier. This may abort the decompensation altogether or shorten its duration (possibly accounting for the fewer hospitalisations seen in PARADIGM-HF). These fewer and shorter episodes of decompensated heart failure result in less sequelae, manifested by a more gradual rise in BNP levels (perhaps explaining the mortality benefit seen in PARADIGM-HF).

Table 1.

DRUG	TRIAL NAME *(patients recruited)*	CONDITION (*study arms)*	FINDINGS *(in treatment arm)*
NESIRITIDE	VMAC *(n=489)*	ADHF *(Nesiritide vs IV GTN vs Placebo)*	• Improved PCWP and all PA pressures within 15 minutes • Improved dysponea at 3 hours
	ASCEND-HF *(n=7141)*	ADHF *(Nesiritide vs Placebo)*	• Signal of improved dyspnoea (not significant) • No 30 day mortality / re-hospitalisation benefit • No effect on urine output • No worsening renal function • More hypotension
	META ANALYSIS: Risk of worsening renal function *(n=1269)*	ADHF *(Nesiritide vs Inotropic / Non-inotropic control)*	• Worsening renal function compared to inotrope / non-inotrope controls • Worsening renal function at any dose of nesiritide • No difference in need for dialysis
ECADOTRIL	U.S. Ecadotril Pilot Safety Study *(n=50)*	CHF (NYHA II-III) *(Ecadotril 50-400 mg vs placebo)*	• Dose-ranging study for 10 weeks • No difference in patient assessed symptoms • No difference in NYHA class • No safety signal raised
	International, Multicentre Ecadotril Dose-ranging Study *(n=279)*	CHF (NYHA II-III) *(Ecadotril 50, 100, 200 and 400mg vs placebo)*	• International dose-ranging study for 13 weeks. • Plasma and urinary cGMP increased • No difference in patient assessed symptoms • No difference in NYHA class • Increased occurrence of aplastic anaemia – clinical development of drug halted.
CANDOXATRIL	Northridge et al. *(n= 60)*	CHF (NYHA I-III) *(Candoxatril vs Captopril vs placebo)*	• Signal of better improvement in exercise tolerance in candoxatril arm at 12 weeks (not significant) • Trend for improved NYHA class and subjective quality of life in both active drug groups (not significant)
	Westheim et al. *(n= 47)*	CHF (NYHA I-II) *(Candoxatril vs Furosemide vs placebo)*	• Candoxatril and Furosemide, compared to placebo, significantly reduce PCWP at day 0 but Candoxatril arm no longer significant at day 42 • Improved cardiac index in both groups at day 0 (Candox > Furo)

			• No change in renin, angiotensin II, aldosterone, noradrenaline activity in Candoxatril arm at day 0 or 42
	Kentsch et al. *(n=24)*	CHF *(Candoxatril vs placebo)*	• Increased plasma cGMP (second messenger of ANP) • Dose dependent increase in PVR & reduction in cardiac index
OMAPATRILAT	IMPRESS *(n=573)*	CHF (NYHA II-IV) *(Omapatrilat vs Lisinopril)*	• Improved NYHA class among NYHA class III & IV patients • Minimal improvement in exercise treadmill test (not significant) • Signal of reduced death or HF admission (not significant)
	OVERTURE *(n=5770)*	CHF (NYHA II-IV) *(Omapatrilat vs Enalapril)*	• Non-inferior to enalapril in preventing death or HF hospitalization requiring IV medication • Less death or all-cause HF hospitalization (post hoc analysis) • 60% relative risk increase in angioedema
	OCTAVE *(n= 25 302)*	Untreated / uncontrolled HPT *(Omapatrilat vs Enalapril)*	• Reduced BP by 3.6/2.0 mmHg more than comparator • Less use of adjunctive anti-hypertensives • More likely to reach BP targets regardless of demographics / comorbids • More frequent angioedema (2.17% vs 0.68%) - including 2 patients with airway compromise
SACUBITRIL / VALSARTAN	PARADIGM-HF *(n=8442)*	CHF (NYHA II-IV & HFrEF: EF ≤ 40%) *(Sacubitril/Valsartan vs Enalapril)*	• 20% RRR in CV death or HF hospitalization • NNT to prevent 1 CV death = 32 • Reduced systolic BP by 3.2 mmHg • Improved subjective quality of life • No significant difference in angioedema rates
	PARAMOUNT *(n=301)*	CHF (NYHA II-III & HFpEF: EF ≥ 45%) *(Sacubitril/Valsartan vs Valsartan)*	• Greater BP reduction - 9.3/4.9 mmHg • Lower NT-proBNP by 12 weeks • LA dimension and volume lower at 36 weeks • No difference ventricular volumes / LVEF / diastolic function • Improved NYHA class at 36 weeks

			• Angioedema only in 1 patient in LCZ arm, nil in valsartan arm
	PARAGON-HF *(currently recruiting)*	CHF (NYHA II-IV & HFpEF: EF ≥ 45%) *(Sacubitril/Valsartan vs Valsartan)*	• Primary outcome: Composite CV death or HF hospitalisation • Secondary outcomes: ○ Cumulative CV death / total HF admissions / nonfatal MI or stroke ○ Change in NYHA class at 8 months ○ Time to AF ○ Time to all cause death

Table 2.

Peptide	Km (μM)	Kcat (min^{-1})	Kcat/Km (min^{-1} μM^{-1})
β-amyloid protein 42	2.8	-	-
Pro-adrenomedullin	6.1	-	-
ANP_{28}	28.3	145	5.1
Substance P	31.9	5062	158.7
Bradykinin	92.2	6364	69.0
BNP_{32}	102	54.3	0.53
Angiotensin II	280	-	-